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EXAMINER	
DUNSTON, JENNIFER ANN	
ART UNIT	**PAPER NUMBER**
1636	

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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/051,481	PIEST ET AL.	
	Examiner	Art Unit	
	Jennifer Dunston	1636	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *06 June 2002*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-26* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-26* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☒ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

Receipt is acknowledged of the declaration and power of attorney, filed 6/6/2002. Claims 1-26 are pending in the instant application.

Priority

Acknowledgment is made of applicant's claim for foreign priority based on an application filed in Europe on 01/19/01. It is noted, however, that applicant has not filed a certified copy of the 01870011.2 application as required by 35 U.S.C. 119(b).

Specification

The abstract of the disclosure is objected to because the abstract contains legal phraseology such as "said donor sequence" (lines 2-3), "said acceptor vector" (line 4), "said donor" (line 6), "said polynucleic acid" (line 7), "said mixture" (line 11), "said second ARFS" (line 14, "said cells" (line 15), "said first ARFS" (line 17, "said polynucleic acid" (line 18), and "said acceptor vector" (line 19). Correction is required. See MPEP § 608.01(b).

Claim Rejections - 35 USC § 112

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-9, 12, 13, and 15-26 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

Claims 1-9, 12, 13, and 15-26 are drawn to a set of compounds that change color in the presence of a first antibiotic resistance functioning sequence. Expression of the antibiotic resistance functioning sequence in a host cell must be capable of providing antibiotic resistance to the cell and catalyzing a color change reaction in the presence of the compound.

The rejected claims thus comprise a set of compounds that undergo a color-change in the presence of any expressed first antibiotic resistance functioning sequence. Thus, the claims encompass an incredibly enormous genus of compounds that must meet specific functional limitations. Given that the range of compounds encompassed by the instant claims is any that is biologically possible, the set of compounds encompassed by the rejected claims is so broad as to be incalculable.

The instant specification describes the use of chromogenic beta-lactamase substrates such as nitrocefin, PADAC or CENTA (e.g. page 22, lines 20-30; Example 6) which change color in the presence of an expressed ampicillin resistance gene. There is no description in the specification as originally filed of any other combination of

antibiotic resistance functioning sequence and compound that would meet such claim limitations.

Even if one accepts that the examples described in the specification meet the claim limitations of the rejected claims with regard to structure and function, the examples are only representative of a few compounds that are cleaved by a single class of antibiotic resistance functioning sequences which provide resistance to beta-lactam antibiotics. The results described are not necessarily predictive of the combinations of chromogenic compounds and antibiotic resistance functioning sequences suitable for counter selection, making it impossible for one to extrapolate from the few examples described herein to those compounds and antibiotic resistance functioning sequences that would necessarily meet the structural/functional characteristics of the rejected claims.

The prior art does not appear to offset the deficiencies of the instant specification in that it does not describe a set of compounds and corresponding antibiotic resistance functioning sequences. Moreover, the prior art does not provide a reliable structural/functional basis for one of skill in the art to envision compounds/sequences that will necessarily meet the structural/functional limitations of the rejected claims to allow growth of bacteria in the presence of the compound and a color change reaction in the presence of an expressed antibiotic functioning sequence.

Therefore, there is no structural/functional basis provided by the prior art or instant specification for one of skill in the art to envision those compounds which result in a visible color change in the presence of an expressed antibiotic resistance

functioning sequence for the incredibly broad genus of compounds and sequences encompassed by the rejected claims. Therefore, one of skill in the art would not have been able to envision a representative number of compounds sufficient to describe the broad genus of chromogenic substrates cleaved by expressed antibiotic resistance functioning sequences encompassed by the rejected claims. One of skill in the art would thus have reasonably concluded that applicants were not in possession of the claimed invention for claims 1-9, 12, 13, and 15-26.

Claims 1-26 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for the use of bacterial cells in the claimed methods, does not reasonably provide enablement for the use of other cell types such as mammalian cells. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims.

Enablement is considered in view of the Wands factors (MPEP 2164.01(A)). These include: nature of the invention, breadth of the claims, guidance of the specification, the existence of working examples, state of the art, predictability of the art and the amount of experimentation necessary. All of the Wands factors have been considered with regard to the instant claims, with the most relevant factors discussed below.

Nature of the invention: The rejected claims are drawn to methods of transferring a polynucleic acid from a donor vector to an acceptor vector where the DNA vectors are

digested by a restriction enzyme(s), the fragments are ligated without purification, the ligated products are transformed into a host cell, and the recombinant vector of interest is selected. The selection step includes growing colonies of the host cell in the presence of an antibiotic, which is a substrate of the antibiotic resistance functioning sequence of the acceptor vector, and a compound that is a chromogenic substrate of the antibiotic resistance functioning sequence of the donor vector. The invention requires the antibiotic resistance functioning sequence to be expressible in the chosen host cell and capable of providing antibiotic resistance.

Breadth of the claims: The claims are broad in that they encompass literally any host cell from any organism.

Guidance of the specification and existence of working examples: The specification asserts that the claimed methods can be practiced with a suitable host cell such as bacteria, specifically *E. coli*, or other host cells (e.g. page 13, lines 12-25). The specific teachings of the specification are all directed towards the use of *E. coli* as a host cell and the ampicillin resistance gene as the first antibiotic functioning sequence. No specific teachings are provided with regard to using any other host cell type.

State of the art: At the time of applicants' invention the art of using the ampicillin resistance gene as a selectable marker in *E. coli* was well developed. Ampicillin, a synthetic derivative of penicillin, functions by interfering with the synthesis of the bacterial cell wall and is primarily effective against gram-positive bacteria but can be effective against certain gram-negative bacteria (Penicillin, Encyclopedia Americana, 2004). There is no art of record to indicate that the ampicillin resistance gene, or other

beta-lactamase encoding genes, would be effective as a selectable marker in other cell types such as mammalian cells.

Predictability of the art: Given the complex nature of the mechanisms by which antibiotics function, and the lack of guidance provided by the specification or prior art with regard to the use of the ampicillin resistance gene in cells other than bacteria, it would have been unpredictable to try and practice the claimed invention with any cell type other than bacterial cells.

Amount of experimentation necessary: The amount of experimentation necessary to carry out the claimed invention is high, as the skilled artisan could not rely on the prior art or the present specification to teach how to use the claimed methods. In order to practice the claimed invention, one of ordinary skill in the art would have to first envision a cell that can be grown in culture and can be transformed with recombinant vectors. Further, one would have to experimentally identify, through trial and error, an antibiotic functioning sequence that meets the following specific functional requirements: (i) it can be functionally expressed, (ii) the expression product provides antibiotic resistance, and (iii) the expression product catalyzes a color change reaction in the presence of a chromogenic substrate. A large number of combinations of antibiotic resistance sequences and compounds would need to be screened before the functional requirements could be met.

Based on the broad scope of the claims, the unpredictability in the art of the invention, the lack of sufficient guidance or working examples in the specification and the quantity of experimentation necessary, it would clearly require undue

experimentation by one of skill in the art to determine how to use the claimed invention. Therefore, the claimed invention of transferring a polynucleic acid from a donor vector to any acceptor vector where the DNA vectors are digested by a restriction enzyme(s), the fragments are ligated without purification, the ligated products are transformed into any host cell, and the recombinant vector of interest is selected is not considered to be fully enabled by the instant specification.

Claims 25 is rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention.

A claim 25 is drawn to the adenoviral adapter vector pIPspAdApt10/Zeo-lacZpart. The exact sequence of this vector is not provided in the instant specification. Although the general method of constructing this vector from starting materials such as pIPspAdApt6 and pClip-LacZ is provided, it is unclear whether the biological starting materials are readily available to the public and will be available over the term of any patent to issue from this application.

The application discloses adenoviral adapter vectors that are encompassed by the definitions for **biological material** set forth in 37 C.F.R. § 1.801. Because it is apparent that this biological material is essential for practicing the claimed invention, it must be obtainable by a reproducible method set forth in the specification or otherwise

be known and readily available to the public as detailed in 37 C.F.R. §§ 1.801 through 1.809.

It is unclear whether this biological material is known and readily available to the public or that the written instructions are sufficient to reproducibly construct this biological material from starting materials known and readily available to the public. Accordingly, availability of such biological material is deemed necessary to satisfy the enablement provisions of 35 U.S.C. § 112. If this biological material is not obtainable or available, the requirements of 35 U.S.C. § 112 may be satisfied by a deposit of the biological material. In order for a deposit to meet all criteria set forth in 37 C.F.R. §§ 1.801-1.809, applicants or assignee must provide assurance of compliance with provisions of 37 C.F.R. §§ 1.801-1.809, in the form of a declaration or applicant's representative must provide a statement. The content of such a declaration or statement is suggested by the enclosed attachment. Because such deposit will not have been made prior to the effective filing date of the instant application, applicant is required to submit a verified statement from a person in a position to corroborate the fact, which states that the biological material which has been deposited is the biological material specifically identified in the application as filed (37 C.F.R. § 1.804). Such a statement need not be verified if the person is an agent or attorney registered to practice before the Office. Applicant is also reminded that the specification must contain reference to the deposit, including deposit (accession) number, date of deposit, name and address of the depository, and the complete taxonomic description. A

statement that all restrictions on the availability to the public of the material so deposited will be irrevocably removed upon granting of a patent is also required.

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 8, 9 and 26 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 8 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. The phrase "wherein said compound includes a negative charge" is unclear in that the referenced compound is a polycationic polymer. The instant specification defines a polycationic polymer as a polymer containing multiple charged subunits containing at least one positive charge and having a net positive charge. The specification does not explicitly define the characteristics of a polycationic polymer which includes a negative charge. It is unclear whether the negative charge is included in the repeating subunits of the polymer, at the terminus of the polymer, as a salt of the polymer or as a counterion in the cross-linking process. It would be remedial to amend the claim language to indicate the structural/functional characteristics of the polycationic polymer encompassed by the term "includes a negative charge."

Claim 9 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. The phrase "wherein said compound includes a positive charge" is unclear in that the referenced compound is a polyanionic polymer. The instant specification defines a polyanionic polymer as a polymer containing multiple charged subunits containing at least one negative charge and having a net negative charge. The specification does not explicitly define the characteristics of a polyanionic polymer which includes a positive charge. It is unclear whether the positive charge is included in the repeating subunits of the polymer, at the terminus of the polymer, as a salt of the polymer or as a counterion in the cross-linking process. It would be remedial to amend the claim language to indicate the structural/functional characteristics of the polyanionic polymer encompassed by the term "includes a positive charge."

Claim 26 is vague and indefinite in that the metes and bounds of the phrase "as defined in claim 25" are unclear. The claim embraces two different statutory classes of invention set forth in 35 U.S.C. 101: i) the "adenoviral vector" is a composition of matter and ii) claim 25 is directed to a process of using an adenoviral vector as an acceptor vector into which polynucleic acid sequence is transferred from a donor vector. It would be remedial to amend the claim language to direct the claimed invention to a single statutory class of invention.

Claim Rejections - 35 USC § 101

35 U.S.C. 101 reads as follows:

> Whoever invents or discovers any new and useful process, machine, manufacture, or composition of matter, or any new and useful improvement thereof, may obtain a patent therefor, subject to the conditions and requirements of this title.

Claim 26 is rejected under 35 U.S.C. 101 because the claimed invention is directed to non-statutory subject matter. The claim is directed to neither a composition of matter nor a process, but rather embraces two different statutory classes of invention set forth in 35 U.S.C. 101, which is drafted so as to set forth the statutory classes of invention in the alternative only.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

The rejected claims are drawn to methods of transferring a polynucleic acid from a donor vector to an acceptor vector where the DNA vectors are digested by a restriction enzyme(s), the fragments are ligated without purification, the ligated products are transformed into bacteria, and the recombinant vector of interest is selected. The selection step includes culturing the bacteria on agar plates in the presence of an antibiotic which is a substrate of the antibiotic resistance gene of the acceptor vector and a chromogenic substrate of the antibiotic resistance gene of the donor vector, resulting in the selection of plasmids containing the acceptor vector backbone without the donor vector backbone. Furthermore, the agar plates can contain polycationic or polyanionic polymers as gelling agents. The phrase "wherein said compound includes a negative charge," which describes a polycationic polymer, is interpreted broadly to include a negative charge present in the repeating subunits of the polymer, at one or both ends of the polymer, as the salt of the polymer or as a counterion in the polymerization reaction. The phrase "wherein said compound includes a positive charge," which describes a polyanionic polymer, is interpreted similarly. The term adenoviral vector is interpreted to include an adenoviral adapter vector as written in claim 24.

Claims 1, 3, 10, 11, 13, 14, 16, 18 and 20 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cohen et al (US Patent No. 4,237,224; see the entire reference) in view of Ferguson et al (Gene, Vol. 16, pages 191-197, 1981; see the

entire reference) further in view of O'Callaghan et al (Antimicrob Agents Chemother. Vol. 1, No. 4, pages 283-288, 1972; see the entire reference).

Cohen et al teach a general method of making molecular chimeras where a foreign DNA and a vector, with a phenotypic property such as antibiotic resistance, are digested with a restriction endonuclease in the same or different vessels, ligated with T4 ligase, transformed into bacteria, and selected by the presence of the phenotypic property (e.g. Column 4, lines 39-62; Column 5, lines 37-53, 59-61; Column 8, lines 27-37). Further, Cohen et al teach that the plasmid chimera may be identified by the phenotypic property of the foreign gene or by characterization of purified DNA by electrophoresis, for example (e.g. Column 8, lines 27-50). The foreign DNA can be obtained from a wide variety of sources such as eukaryotic cells, prokaryotic cells, viruses and bacteriophage and can include one or more genes or operons (e.g. Column 5, lines 59-65). Moreover, Cohen et al teach that the recipient DNA may be any molecule capable of replication in bacteria such as viruses. During a subcloning procedure, Cohen et al teach heat-inactivation of restriction enzymes such as EcoRI (e.g. Example I).

Cohen et al do not provide working examples demonstrating the transfer of a polynucleic acid sequence from a donor vector with an ampicillin resistance gene to an acceptor vector.

Ferguson et al teach a method of subcloning from a vector such as YRp7, which carries an ampicillin resistance gene, to subcloning vectors such as pRC1, pRC2 or pRC3, which carry a kanamycin resistance gene (e.g. Introduction, page 192). To

demonstrate the utility of the subcloning procedure, Ferguson et al transferred the yeast *CDC28* gene from YRp7 (a clone that was identified as part of a yeast genomic DNA library) into pRC1 by digesting the DNA with Sau3A, ligating the restriction fragments overnight with T4 ligase, transforming *E. coli*, and selecting for transformants on LB plus kanamycin. Ferguson et al differentiated the insertion of the *CDC28* gene from the insertion of the YRp7 plasmid backbone by testing for complementation of the *cdc28-4*^{ts} mutant phenotype in yeast by the expression of the *CDC28* gene from the pRC1 plasmid (e.g. (c) Subcloning of a DNA fragment containing the yeast *CDC28* gene using pRC1, pages 195-196). Transfer of the gene from a vector with ampicillin resistance to that with kanamycin resistance allowed selection on kanamycin and the elimination of the Yrp7 parent plasmid which escaped Sau3A digestion (e.g. Discussion, page 196).

Cohen et al and Ferguson et al do not explicitly teach the use of a chromogenic compound for the phenotypic selection of recombinant acceptor vector containing the ampicillin resistance gene of the donor vector.

O'Callaghan et al teach a compound, 3-(2,4-dinitrostyryl)-(6R,7R)-7-(2-thienylacetamido)-ceph-3-em-4-carboxylic acid, E-isomer (a.k.a. nitrocefin or 87/312), which undergoes a distinctive color change when hydrolyzed by beta-lactamases (e.g. Introduction). Further, O'Callaghan et al teach a method of using nitrocefin to detect beta-lactamase expression in bacterial colonies by dropping a solution of nitrocefin onto bacterial colonies growing on agar plates (e.g. Materials and Methods section; Results section). The color change reaction taught by O'Callaghan et al was almost immediate with powerful beta-lactamase producers and could be detected after a 30 min

incubation with the weak producers, making the assay sensitive and absent of false-negative results (e.g. see Results and Discussion, page 288).

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Cohen et al to include the specific examples taught by Ferguson et al, because Cohen et al teach a general method for the production of molecular chimeras. Cohen et al teach it is within the skill of the art to use a variety of molecules as foreign DNA and vector DNA component in the method. Further, Ferguson et al teach the use of plasmids with two different antibiotic functioning sequences to eliminate the parent (i.e. donor) plasmid. Moreover, it would have been obvious to one of ordinary skill in the art to utilize the chromogenic compound taught by O'Callaghan et al, because Cohen et al and Ferguson et al teach it is within the skill of the art to use a phenotypic property, such as antibiotic resistance, to identify the insertion of a foreign DNA sequence. The skilled artisan would have been motivated to make such modifications to the method of Cohen et al in order to receive any of the expected benefits recited by Ferguson et al, with regard to the ability to transfer a DNA insert from one vector to another while simultaneously eliminating the parent plasmid, and O'Callaghan et al, with regard to the detection of bacterial beta-lactamase expression in the absence of false-negative results. Absent any evidence to the contrary, there would have been a reasonable expectation of success in modifying the teachings of Cohen et al to include a number of different donor vectors and acceptor vectors and the use of a phenotypic marker to select against the insertion of vector

backbone sequences such as the chromogenic compound and taught by O'Callaghan et al.

Claims 1, 2, 10, 13, 14, 16 and 18 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cohen et al in view of Ferguson et al further in view of Schindler et al. (US Patent No. 4,353,824; see the entire reference).

The teachings of each of the Cohen et al and Ferguson et al references are described above and are applied as before, except:

Cohen et al and Ferguson et al do not explicitly teach the addition of a chromogenic compound to the media for the phenotypic selection of bacteria containing recombinant acceptor vector containing the ampicillin resistance gene of the donor vector.

Schindler et al teach chromogenic beta-lactamase substrates which are characterized by a distinctly visible color change and are suitable for the detection of bacteria producing beta-lactamase (e.g. Abstract; Column 1, lines 42-50). The compounds taught by Schindler et al are stable to hydrolytic degradation in a temperature range from –180 °C to 60 °C in solutions suitable for the activity of the beta-lactamases and the viability of bacteria (Column 2, lines 1-19). The compounds can be diluted in hot agar-agar solution containing all nutrients necessary for the growth of bacteria and the color change can be detected in the neighborhood of the bacterial colonies expressing beta-lactamase (e.g. Column 9, lines 6-17; Column 10, lines 1-5).

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Cohen et al and Ferguson et al to include the chromogenic substrates taught by Schindler et al because Cohen et al teach the use of phenotypic properties to determine the identity of the foreign DNA in the recombinant molecule. The skilled artisan would have been motivated to make such a modification in order to receive the expected benefits recited in the teachings of Schindler et al with regard to the ability to add the chromogenic substrate directly to media capable of supporting the growth of bacteria and the identification of the ampicillin resistance gene in the recombinant molecule. Absent any evidence to the contrary, there would have been a reasonable expectation of success in modifying the teachings Cohen et al and Ferguson et al to include the chromogenic beta-lactamase substrates taught by Schindler et al.

Claims 4, 5, 7 and 8 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cohen et al and Ferguson et al in view of Schindler et al and further in view of Vorlop et al (Methods in Enzymology, Vol. 135, pages 259-268, 1987; see the entire reference).

The teachings of each of the Cohen et al, Ferguson et al and Schindler et al references are described above and are applied as before, except:

Cohen et al, Ferguson et al and Schindler et al teach the selection of bacteria containing recombinant molecules on agar plates. Cohen et al, Ferguson et al and

Schindler et al do not teach the addition of a charged polymer gelling agent to the growth medium.

Vorlop et al teach a general method for the entrapment of whole cells in chitosan, which is a readily available commercial polycation (e.g. page 259-260). Further, the chitosan can be cross-linked with low-molecular-weight counterions such as pyrophosphate, tripolyphosphate, tetrapolyphosphate, octapoplyphosphate or hexametaphosphate, for example (e.g. page 260; Table I). The method taught by Vorlop et al includes the immobilization of living cells such as *E. coli* (e.g. page 267) by suspending up to 8 g of wet cells in 82 ml of chitosan-acetate solution, adding the suspension drop wise into 500 ml of gently stirred 1.5% polyphosphate solution (pH 5.7) in an immobilization apparatus, decanting the polyphosphate solution after a 30 min incubation, washing the chitosan beads with 0.1 M phosphate buffer (pH 7.5), and incubating in a growth medium (e.g. page 262 and 267). Vorlop et al teach the use of cells entrapped in the chitosan matrix to catalyze reactions such as the formation of 6-APA from penicillin G (e.g. page 266). The entrapment of cells within the polymer results in biocatalysts with good mechanical properties, high cell loading, and high activity (e.g. page 259).

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Cohen et al, Ferguson et al and Schindler et al to include the charged polymer gelling agent taught by Vorlop et al, because the authors teach the use of phenotypic properties to select the presence of recombinant molecules in bacteria where the identification of the phenotypic properties occurs on a gelled

media such as agar. The skilled artisan would have been motivated to make such a modification in order to receive the expected benefits of good mechanical properties, high cell loading and high activity taught by Vorlop et al. Absent any evidence to the contrary, there would have been a reasonable expectation of success in modifying the teachings of Cohen et al, Ferguson et al and Schindler et al to include the charged polymer gelling agent taught by Vorlop et al.

Claims 6 and 9 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cohen et al and Ferguson et al in view of Schindler et al further in view of Chibata et al (Methods in Enzymology, Vol. 135, pages 189-198; see the entire reference).

The teachings of each of the Cohen et al, Ferguson et al and Schindler et al references are described above and are applied as before, except:

Cohen et al, Ferguson et al and Schindler et al teach the selection of bacteria containing recombinant molecules on agar plates. Cohen et al, Ferguson et al and Schindler et al do not teach the addition of a polyanionic gelling agent to the growth medium.

Chibata et al teach the use of the polyanionic molecule κ-carrageenan to immobilize microbial cells by dissolving κ-carrageenan in warmed physiological saline at a concentration of 3.4% (w/v) and forming a gel by cooling and/or contacting with positively charged ions or organic solvents (e.g. page 190-191; Table I). Chibata et al teach the preferred method of using cooling and/or contact with K^+ or NH_4^+ to form a κ-

carrageenan gel suitable for entrapping microbial cells and growing them in a nutrient medium (page 194). Further, Chibata et al report high enzyme activities in κ-carrageenan gels of various shapes formed by contact with K^+ ions (e.g. page 197-198).

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Cohen et al, Ferguson et al and Schindler et al to include the charged polymer gelling agent taught by Chibata et al, because the authors teach the use of phenotypic properties to select the presence of recombinant molecules in bacteria where the identification of the phenotypic properties occurs on a gelled media such as agar. The skilled artisan would have been motivated to make such a modification in order to receive the expected benefits of high enzyme activity of proteins expressed by the bacteria and the ability to tailor the shape of the gel to suit the specific application. Absent any evidence to the contrary, there would have been a reasonable expectation of success in modifying the teachings of Cohen et al, Ferguson et al and Schindler et al to include the charged polymer gelling agent taught by Chibata et al.

Claim 15 is rejected under 35 U.S.C. 103(a) as being unpatentable over Cohen et al and Ferguson et al in view of Schindler et al further in view of the Invitrogen product manual for catalog numbers V850-01 and V855-01 (copyright 1998-2001; see the entire reference).

The teachings of each of the Cohen et al, Ferguson et al and Schindler et al references are described above and are applied as before, except:

The Ferguson et al reference specifically exemplifies the use of an acceptor vector which carries a kanamycin resistance gene. The Ferguson et al reference does not explicitly teach the use of an acceptor vector with a functioning zeocin resistance gene.

The Invitrogen product manual for catalog numbers V850-01 and V855-01 teaches plasmid vectors, pZeoSV2 (+/-) and pZeoSV3/lacZ, which do not encode an ampicillin resistance gene and can be used in standard subcloning procedures (e.g. pages 6, 8, 12). The insertion of foreign DNA sequences into the polylinker of either vector allows constitutive mammalian expression of the gene of interest (e.g. page 9).

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Cohen et al, Ferguson et al and Schindler et al to include the use of the Invitrogen plasmids containing a zeocin resistance gene, because Cohen et al teach it is within the skill of the art to use a number of different acceptor molecules for the creation of chimeric molecules, and the plasmids taught by Invitrogen lack the ampicillin resistance gene which makes them compatible with the selection systems taught by Ferguson et al and Schindler et al. The skilled artisan would have been motivated to make such a modification in order to receive any of the expected benefits recited in the teachings of the Invitrogen product manual with regard to using the pZeoSV2 (+/-) or pZeoSV3/lacZ vectors such as the ability to constitutively express a foreign DNA in a mammalian cell. Absent any evidence to the contrary, there would have been a reasonable expectation of success in modifying the teachings of Cohen et al, Ferguson et al and Schindler et al to include the pZeoSV2 (+/-) and pZeoSV3/lacZ

plasmids taught by the Invitrogen product manual for catalog numbers V850-01 and V855-01.

Claim 18 is rejected under 35 U.S.C. 103(a) as being unpatentable over Cohen et al and Ferguson et al in view of Schindler et al further in view of Cortese (The Scientist, Vol. 14, No. 11, page 26, 2000; see the entire reference).

The teachings of each of the Cohen et al, Ferguson et al and Schindler et al references are described above and are applied as before, except:

Cohen et al, Ferguson et al and Schindler et al use standard molecular biology techniques for the production and selection of recombinant molecules. Cohen et al, Ferguson et al and Schindler et al do not teach the use of high-throughput techniques to obtain recombinant molecules.

Cortese teaches robots that array large DNA libraries and automatic colony pickers that can transfer genetic material from growth plates to multiwell microplates to reduce the workload (e.g. Helping Hands section). Furthermore, Cortese teaches the availability of these tools from multiple companies.

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Cohen et al, Ferguson et al and Schindler et al to include the automated machinery taught by Cortese, because the procedures taught by Cohen et al, Ferguson et al and Schindler et al are amenable to automation. The skilled artisan would have been motivated to make such a modification in order to receive the

expected benefits of the ability to handle large numbers of samples in an automated fashion to reduce the workload. Absent any evidence to the contrary, there would have been a reasonable expectation of success in modifying the teachings of Cohen et al, Ferguson et al and Schindler et al to include the automated machinery taught by Cortese.

Claims 12, 21, 23 and 24 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cohen et al and Ferguson et al in view of Schindler et al and further in view of He et al (US Patent No. 5,922,576; see the entire reference).

The teachings of each of the Cohen et al, Ferguson et al and Schindler et al references are described above and are applied as before, except:

The Ferguson et al reference exemplifies the use of yeast expression plasmids. The Ferguson et al reference does not explicitly teach the use of an adenoviral expression vector.

He et al teach a method for generating recombinant adenoviral vectors which, in one embodiment, includes the use of two bacterial plasmids: 1) a plasmid such as pShuttle-CMV that comprises a restriction enzyme site for insertion of a desired gene and sequences which allow recombination with adenoviral genomic DNA and 2) a plasmid such as pAdEasy-1 that contains an adenoviral genome and is capable of undergoing homologous recombination with the first plasmid (e.g. Abstract; Column 1, lines 65-67; Column 2, lines 24-42). He et al teach the insertion of genes into the

plasmids by restriction enzyme cleavage and ligation (e.g. Column 4, lines 17-19). The pShuttle-CMV vector taught by He et al contains a kanamycin resistance gene, left and right inverted terminal repeats (ITR), a CMV promoter, polylinker and polyadenylation signal and lacks E1 (e.g. Column 8, lines 40-62; Figure 2). This vector also includes part of the E2B region in that "part" can be defined as a single nucleotide. The use of the system provided by He et al expedites the process of generation and testing recombinant adenoviruses (e.g. Abstract).

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Cohen et al, Ferguson et al and Schindler et al to include the vectors taught by He et al as acceptor vectors, because Cohen et al teach it is within the skill of the art to use a number of different acceptor molecules for the creation of chimeric molecules. Further, vectors taught by He et al (e.g. pShuttle-CMV) lack an ampicillin resistance gene which makes them compatible with the selection systems taught by Ferguson et al and Schindler et al. The skilled artisan would have been motivated to make such a modification in order to receive the expected benefits recited by He et al with regard to the use of such plasmids to generate recombinant adenoviruses.

1, 2, 10, 13, 14, 17 and 18 are rejected under 35 U.S.C. 103(a) as being unpatentable over Kirschman et al (Gene, Vol. 68, No. 1, pages 163-165, 1988; see the entire reference) in view of O'Callaghan et al.

Kirschman et al teach a strategy for expediting the re-cloning of large numbers of inserts using a recipient vector with an alternative selectable marker relative to the donor vector (e.g. Introduction). Kirschman et al teach the transfer of polynucleic sequences from a maize genomic library in pUC12 (a plasmid which carries and ampicillin resistance gene) to plasmid pJKKmf(-), which has a kanamycin resistance gene (e.g. see (a) Construction of plasmids pJKKmf(-) and pJKSp/Smf(-) and (b) Rapid subcloning with pJKKmf(-)). The strategy taught by Kirschman et al provides a method of digesting a library of plasmids and pJKKmf(-) with PstI, mixing the digested plasmids in approximately equimolar amounts without prior purification, ligating and transforming the recombinant plasmids into *E. coli*. Further, Kirschman et al teach counter selection of recombinant plasmids containing the ampicillin resistance gene rather than the desired PstI fragment by selecting colonies that were sensitive to ampicillin.

The Kirschman et al reference does not explicitly teach the use of a chromogenic substrate as a compound for the counter selection of recombinant plasmids containing the ampicillin resistance gene.

O'Callaghan et al teach a compound, 3-(2,4-dinitrostyryl)-(6R,7R)-7-(2-thienylacetamido)-ceph-3-em-4-carboxylic acid, E-isomer (a.k.a. nitrocefin or 87/312), which undergoes a distinctive color change when hydrolyzed by beta-lactamases (e.g. Introduction). Further, O'Callaghan et al teach a method of using nitrocefin to detect beta-lactamase expression in bacterial colonies by dropping a solution of nitrocefin onto bacterial colonies growing on agar plates (e.g. Results section). The color change reaction taught by O'Callaghan et al was almost immediate with powerful beta-

lactamase producers and could be detected after a 30 min incubation with the weak producers, making the assay sensitive and absent of false-negative results (e.g. see Results and Discussion, page 288).

It would have been obvious to one of ordinary skill in the art at the time of the invention to modify the teachings of Kirschman et al to include the chromogenic compound taught by O'Callaghan et al, because Kirschman et al teach it is within the skill of the art to use a compound for the detection of an expressed ampicillin resistance gene, and O'Callaghan et al teach the detection of beta-lactamase expression in bacteria grown on agar plates. The skilled artisan would have been motivated to make such a modification in order to receive any of the expected benefits recited by O'Callaghan et al with regard to the detection of bacterial beta-lactamase expression in the absence of false-negative results. Absent any evidence to the contrary, there would have been a reasonable expectation of success in modifying the teachings of Kirschman et al to include the chromogenic compound taught by O'Callaghan et al.

Conclusion

No claims are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Jennifer Dunston whose telephone number is 571-272-2916. The examiner can normally be reached on M-F, 9 am to 5 pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Remy Yucel can be reached on 571-272-0781. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

Jennifer Dunston
Examiner
Art Unit 1636

jad

GERRY LEFFERS
PRIMARY EXAMINER

SUGGESTION FOR DEPOSIT OF BIOLOGICAL MATERIAL

A declaration by applicant or assignee, or a statement by applicant's agent identifying a deposit of biological material and averring the following may be sufficient to overcome an objection or rejection based on a lack of availability of biological material. Such a declaration:

1. Identifies declarant.

2. States that a deposit of the material has been made in a depository affording permanence of the deposit and ready accessibility thereto by the public if a patent is granted. The depository is to be identified by name and address. (See 37 C.F.R. § 1.803).

3. States that the deposited material has been accorded a specific (recited) accession number.

4. States that all restrictions on the availability to the public of the material so deposited will be irrevocably removed upon the granting of the patent. (See 37 C.F.R. § 1.808(a)(2)).

5. States that the material has been deposited under conditions that assure that access to the material will be available during the pendency of the patent application to one determined by the Commissioner to be entitled thereto under 37 C.F.R. § 1.14 and 35 U.S.C. § 122. (See 37 C.F.R. § 1.808(a)(1)).

6. States that the deposited material will be maintained with all the care necessary to keep it viable and uncontaminated for a period of at least five years after the most recent request for the furnishing of a sample of the deposited microorganism, and in any case, for a period of at least thirty (30) years after the date of deposit or for the enforceable life of the patent, whichever period is longer. See 37 C.F.R. § 1.806).

7. That he/she declares further that all statements made therein of his/her own knowledge are true and that all statements made on information and belief are believed to be true; and further, that these statements were made with knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the instant patent application or any patent issuing thereon.

Alternatively, it may be averred that deposited material has been accepted for deposit under the Budapest Treaty on the International Recognition of the Deposit of Microorganisms for the Purposes of Patent Procedure (e.g., see 961 OG 21, 1977) and that all restrictions on the availability to the public of the material so deposited will be irrevocably removed upon the granting of a patent.

Additionally, the deposit must be referred to in the body of the specification and be identified by deposit (accession) number, date of deposit, name and address of the depository, and the complete taxonomic description.